

AD _____

GRANT NUMBER DAMD17-94-J-4011

TITLE: Ethanol Disruption of Synaptic Neurotransmission

PRINCIPAL INVESTIGATOR: Robert S. Aronstam, Ph.D.

CONTRACTING ORGANIZATION: Guthrie Research Institute
Sayre, Pennsylvania 18840

REPORT DATE: April 1998

TYPE OF REPORT: Final

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for public release;
distribution unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

DTIC QUALITY INSPECTED 1

19980617 066

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

Public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden, to Washington Headquarters Services, Directorate for Information Operations and Reports, 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302, and to the Office of Management and Budget, Paperwork Reduction Project (0704-0188), Washington, DC 20503.

1. AGENCY USE ONLY (Leave blank)		2. REPORT DATE April 1998		3. REPORT TYPE AND DATES COVERED Final (21 Mar 94 - 20 Mar 98)	
4. TITLE AND SUBTITLE Ethanol Disruption of Synaptic Neurotransmission				5. FUNDING NUMBERS DAMD17-94-J-4011	
6. AUTHOR(S) Aronstam, Robert S., Ph.D.					
7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES) Guthrie Research Institute Sayre, Pennsylvania 18840				8. PERFORMING ORGANIZATION REPORT NUMBER	
9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES) U.S. Army Medical Research and Materiel Command Fort Detrick, Maryland 21702-5012				10. SPONSORING / MONITORING AGENCY REPORT NUMBER	
11. SUPPLEMENTARY NOTES					
12a. DISTRIBUTION / AVAILABILITY STATEMENT Approved for public release; distribution unlimited				12b. DISTRIBUTION CODE	
13. ABSTRACT (Maximum 200 words) The goal of this research program was to understand how acute and chronic ethanol administration disrupts synaptic transmission in the central nervous system. The overall hypothesis was that <i>ethanol depresses neurotransmission at neurotransmitter receptors by disrupting receptor-G protein interactions</i> . To this end, the influence of ethanol was measured on receptor-ligand binding (including the guanine nucleotide sensitivity of agonist binding), receptor control of the G protein cycle, and receptor regulation of specific signal transduction processes. Signal transduction processes studied included stimulation of phosphatidylinositol metabolism, release of arachidonic acid, control of adenylate cyclase, alteration of intracellular calcium concentration, and cell-cell adhesion. Chronic effects of ethanol on synaptic signaling processes, including receptor and G protein expression, were studied in brain tissue from rats treated with ethanol for 8-9 weeks, as well as in cell cultures treated for 48 hours. To gain a better understanding of the molecular action of ethanol on control of receptor expression, the promoters regions of muscarinic receptor genes were analyzed in detail. These studies revealed numerous specific actions of ethanol on synaptic signaling pathways. It is clear, however, that receptors differ in their sensitivity to ethanol and that system must be characterized separately.					
14. SUBJECT TERMS Ethanol; G proteins; signal transduction; neurotransmitter receptors				15. NUMBER OF PAGES 373	
				16. PRICE CODE	
17. SECURITY CLASSIFICATION OF REPORT Unclassified	18. SECURITY CLASSIFICATION OF THIS PAGE Unclassified	19. SECURITY CLASSIFICATION OF ABSTRACT Unclassified	20. LIMITATION OF ABSTRACT Unlimited		

FOREWORD

Opinions, interpretations, conclusions and recommendations are those of the author and are not necessarily endorsed by the U.S. Army.

✓ Where copyrighted material is quoted, permission has been obtained to use such material.

✓ Where material from documents designated for limited distribution is quoted, permission has been obtained to use the material.

✓ Citations of commercial organizations and trade names in this report do not constitute an official Department of Army endorsement or approval of the products or services of these organizations.

✓ In conducting research using animals, the investigator(s) adhered to the "Guide for the Care and Use of Laboratory Animals," prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Resources, National Research Council (NIH Publication No. 86-23, Revised 1985).

 For the protection of human subjects, the investigator(s) adhered to policies of applicable Federal Law 45 CFR 46.

 In conducting research utilizing recombinant DNA technology, the investigator(s) adhered to current guidelines promulgated by the National Institutes of Health.

 In the conduct of research utilizing recombinant DNA, the investigator(s) adhered to the NIH Guidelines for Research Involving Recombinant DNA Molecules.

 In the conduct of research involving hazardous organisms, the investigator(s) adhered to the CDC-NIH Guide for Biosafety in Microbiological and Biomedical Laboratories.

Robert S. Aroustam

PI - Signature

4/19/98
Date

Table of Contents

1. Front Cover	1
2. SF 298 Report Documentation Page	2
3. Foreword	3
4. Table of Contents	4
5. Introduction	5
6. Body	8
A. Background	8
B. Ligand Binding Studies.....	14
1. Muscarinic Receptors.....	14
2. Mu Opiate Receptors	29
3. Other Receptors	32
C. G Protein Studies.....	33
D. Signal Transduction Studies	41
1. Adenylate Cyclase Studies	41
2. Ca ²⁺ Response Studies.....	46
3. Cell Adhesion Studies	69
E. Chronic Treatment Studies	71
1. G Protein Expression.....	71
2. Signal Transduction.....	79
a. Arachidonic acid	79
b. Phosphoinositide metabolism.....	88
F. Molecular Analysis of Muscarinic Receptors.	100
7. Conclusions	124
8. Bibliography: Publications Supported by this Grant....	127
9. Personnel supported by this Grant	130